#### **End of Result Set**

Generate Collection Print

5482846

File: USPT

L1: Entry 1 of 1

Jun 29, 1999

US-PAT-NO: 5916787

DOCUMENT-IDENTIFIER: US 5916787 A

TITLE: Ethanol production in gram-positive microbes

DATE-ISSUED: June 29, 1999

INVENTOR - INFORMATION:

NAME CITY STATE ZIP CODE COUNTRY

Ingram; Lonnie O'Neal Gainesville FL Barbosa-Alleyne; Maria D. F. Gainesville FL

US-CL-CURRENT: 435/161; 257/E21.317, 435/162, 435/163, 435/165, 435/252.31, 435/320.1

#### CLAIMS:

#### We claim:

- 1. A Gram-positive bacterium which has been transformed with heterologous genes encoding alcohol dehydrogenase and pyruvate decarboxylase wherein said genes are expressed at sufficient levels to confer upon said Gram-positive bacterium transformant the ability to produce ethanol as a fermentation product.
- 2. The Gram-positive bacterium, according to claim 1, wherein said host is selected from the group consisting of Bacillus, Lactobacillus, Streptococcus, Fibribacter, Ruminococcus, Pediococcus, Cytophaga, Cellulomonas, Bacteroides, and Clostridium.
- 3. The Gram-positive bacterium according to claim 2, wherein said host is a Bacillus  ${\tt sp.}$
- 4. The Gram-positive bacterium, according to claim 3, wherein said Bacillus sp. is selected from the group consisting of B. subtilis and B. polymyxa.
- 5. The Gram-positive bacterium, according to claim 1, which has been transformed with Z. mobilis genes encoding alcohol dehydrogenase and pyruvate decarboxylase.
- 6. The Gram-positive bacterium according to claim 1, wherein said bacterium is further transformed with a gene encoding an enzyme which degrades oligosaccharides.
- 7. The Gram-positive bacterium, according to claim 6, wherein said enzyme which degrades oligosaccharides is a polysaccharase.
- 8. The Gram-positive bacterium according to claim 7, wherein said polysaccharase is selected from the group consisting of cellulolytic, xylanolytic, and starch-degrading enzymes.
- 9. The Gram-positive bacterium, according to claim 1, wherein said heterologous genes are incorporated onto the chromosome of said bacterium.

- 10. A method for the production of ethanol, said method comprising transforming a Gram-positive bacterial host with heterologous genes encoding pyruvate decarboxylase and alcohol dehydrogenase wherein said genes are expressed at sufficient levels to result in the production of ethanol as a fermentation product.
- 11. The method, according to claim 10, wherein said host is selected from the group consisting of Bacillus, Lactobacillus, Streptococcus, Fibribacter, Ruminococcus, Pediococcus, Cytophaga, Cellulomonas, Bacteroides, and Clostridium.
- 12. The method, according to claim 11, wherein said host is a Bacillus sp.
- 13. The method, according to claim 12, wherein said Bacillus sp. is selected from the group consisting of B. subtilis and B. polymyxa.
- 14. The method, according to claim 10, wherein said Gram-positive bacterium has been transformed with Z. mobilis genes encoding alcohol dehydrogenase and pyruvate decarboxylase.
- 15. The method, according to claim 10, wherein said bacterium is further transformed with a gene encoding an enzyme which degrades oligosaccharides.
- 16. The method, according to claim 15, wherein said enzyme which degrades oligosaccharides is a polysaccharase.
- 17. A method for reducing the accumulation of acidic metabolic products in the growth medium of Gram-positive bacteria, said method comprising transforming said bacteria with heterologous genes which express alcohol dehydrogenase and pyruvate decarboxylase at sufficient levels to result in the production of ethanol as a fermentation product.
- 18. A plasmid designated pLOI1500.

#### **End of Result Set**

Generate Collection Print

L3: Entry 1 of 1

File: USPT

Mar 19, 1991

US-PAT-NO: 5000000

DOCUMENT-IDENTIFIER: US 5000000 A

TITLE: Ethanol production by Escherichia coli strains co-expressing Zymomonas PDC and

ADH genes

DATE-ISSUED: March 19, 1991

INVENTOR-INFORMATION:

NAME CITY STATE ZIP CODE COUNTRY

Ingram; Lonnie O. Gainesville FL
Conway; Tyrrell Lincoln NE
Alterthum; Flavio Gainesville FL

US-CL-CURRENT: 435/161; 257/E21.317, 435/170, 435/252.3, 435/252.33, 435/320.1,

<u>435/488</u>

#### CLAIMS:

#### We claim:

- 1. An Escherichia coli, which has been transformed with Zymomonas mobilis genes coding for alcohol dehydrogenase and pyruvate decarboxylase wherein said genes are expressed at sufficient levels to confer upon said Escherichia coli transformant the ability to produce ethanol as a fermentation product.
- 2. The Escherichia coli, according to claim 1, wherein the Escherichia coli, prior to transformation, is selected from the group consisting of ATCC 8677, ATCC 8739, ATCC 9637, ATCC 11303, ATCC 11775, ATCC 14948, ATCC 15224, and ATCC 23227.
- 3. The Escherichia coli, according to claim 1, wherein said Escherichia coli has been transformed with a plasmid selected from the group consisting of pLOI308-10, pLOI297, and pLOI308-11.
- 4. The Escherichia coli, according to claim 3, wherein said Escherichia coli has been transformed with pLOI297.
- 5. A method for the production of ethanol, said method comprising transforming an Escherichia coli with Zymomonas mobilis genes coding for pyruvate decarboxylase and alcohol dehydrogenase wherein said genes are expressed by the transformed Escherichia coli at sufficient levels to result in the production of ethanol as a fermentation product when said Escherichia coli is grown in an appropriate medium.
- 6. The method, according to claim 5, wherein said Escherichia coli is transformed with a plasmid selected from the group consisting of pLOI308-10, pLOI297, and pLOI308-11.
- 7. The method, according to claim 6, wherein said Escherichia coli has been transformed with pLOI297.

Generate Collection Print

L2: Entry 1 of 6

File: USPT

Dec 25, 2001

US-PAT-NO: 6333181

DOCUMENT-IDENTIFIER: US 6333181 B1

TITLE: Ethanol production from lignocellulose

DATE-ISSUED: December 25, 2001

INVENTOR-INFORMATION:

NAME CITY STATE ZIP CODE COUNTRY

Ingram; Lonnie O. Gainesville FL Wood; Brent E. Gainesville FL

US-CL-CURRENT: 435/165; 435/163, 435/170, 435/173.2, 435/173.8, 435/277, 435/278,

435/72, 435/99

#### CLAIMS:

What is claimed is:

- 1. A method for producing ethanol from lignocellulose comprising discontinuously treating an aqueous mixture containing lignocellulose, a cellulase and an ethanologenic microorganism with ultrasound under conditions sufficient for hydrolysis of said lignocellulose to occur, to thereby produce ethanol.
- 2. The method according to claim 1 wherein said cellulase is provided by a cellulase-producing microorganism in said aqueous mixture.
- 3. The method according to claim 1 wherein said aqueous mixture is treated with ultrasound at a frequency of between about 2 and 200 kHz.
- 4. The method according to claim 1 wherein said ethanologenic microorganism is an ethanologenic bacteria or yeast.
- 5. The method according to claim 4 wherein said ethanologenic microorganism is a bacteria or yeast which expresses one or more enzymes which, individually or together, convert a sugar to ethanol.
- 6. The method according to claim 4 wherein said ethanologenic microorganism expresses enzymes which, individually or together, convert pentose and hexose to ethanol.
- 7. The method according to claim 4 wherein said ethanologenic microorganism expresses alcohol dehydrogenase and pyruvate decarboxylase.
- 8. The method according to claim 7 wherein said alcohol dehydrogenase and pyruvate decarboxylase are from Zymomonas mobilis.
- 9. The method according to claim 4 wherein said ethanologenic microorganism expresses xylose isomerase, xylulokinase, transaldolase, and transketolase.
- 10. The method according to claim 9 wherein said xylose isomerase, xylulokinase, transaldolase, and transketolase are from Escherichia coli.

- 11. The method according to claim 9 wherein said xylose isomerase, xylulokinase, transaldolase, and transketolase are from Klebsiella oxytoca.
- 12. The method according to claim 9 wherein said xylose isomerase, xylulokinase, transaldolase, and transketolase are from Erwinia species.
- 13. The method according to claim 4 wherein said ethanologenic microorganism expresses alcohol dehydrogenase, pyruvate decarboxylase, xylose isomerase, xylulokinase, transaldolase, and transketolase.
- 14. The method according to claim 13 wherein said ethanologenic microorganism is a recombinant microorganism expressing Zymomonas mobilis alcohol dehydrogenase and pyruvate decarboxylase wherein said microorganism is selected from the group consisting of Escherichia coli, Klebsiella oxytoca, and Erwinia species.
- 15. The method according to claim 14 wherein said ethanologenic microorganism is Klebsiella oxytoca P2.
- 16. The method according to claim 14, wherein said ethanologenic microorganism is Escherichia coli KO11.

Generate Collection

**Print** 

**Search Results** - Record(s) 1 through 6 of 6 returned.

☐ 1. Document ID: US 6333181 B1

L2: Entry 1 of 6

File: USPT

Dec 25, 2001

US-PAT-NO: 6333181

DOCUMENT-IDENTIFIER: US 6333181 B1

TITLE: Ethanol production from lignocellulose

DATE-ISSUED: December 25, 2001

INVENTOR-INFORMATION:

NAME

· CITY

STATE

ZIP CODE

COUNTRY

Ingram; Lonnie O.

Gainesville

FL

Wood; Brent E.

Gainesville

 $\mathtt{FL}$ 

US-CL-CURRENT: <u>435/165</u>; <u>435/163</u>, <u>435/170</u>, <u>435/173.2</u>, <u>435/173.8</u>, <u>435/277</u>, <u>435/278</u>, <u>435/72</u>, <u>435/99</u>

Full Title Citation Front Review Classification Date Reference Sequences Attachments Claims KWIC Draw. Description

☐ 2. Document ID: US 6280986 B1

L2: Entry 2 of 6

File: USPT

Aug 28, 2001

US-PAT-NO: 6280986

DOCUMENT-IDENTIFIER: US 6280986 B1

TITLE: Stabilization of pet operon plasmids and ethanol production in bacterial strains lacking lactate dehydrogenase and pyruvate formate lyase activities

DATE-ISSUED: August 28, 2001

INVENTOR-INFORMATION:

NAME CITY STATE ZIP CODE COUNTRY

Hespell; Robert B. late of Peoria IL
Wyckoff; Herbert A. Roscoe IL
Dien; Bruce S. Peoria IL
Bothast; Rodney J. East Peoria IL

US-CL-CURRENT: 435/161; 435/252.3, 435/252.33, 435/320.1

Full Title Citation Front Review Classification Date Reference Sequences Attachments Claims KMC Draw. Desc Image ☐ 3. Document ID: US 6130076 A

L2: Entry 3 of 6

File: USPT

Oct 10, 2000

US-PAT-NO: 6130076

DOCUMENT-IDENTIFIER: US 6130076 A

TITLE: Ethanol production using a soy hydrolysate-based medium or a yeast

autolysate-based medium

DATE-ISSUED: October 10, 2000

INVENTOR-INFORMATION:

NAME CIT

CITY

STATE ZIP CODE

COUNTRY

Ingram; Lonnie O.

Gainesville

FL

US-CL-CURRENT: 435/161

Full	Title	Citation	Front	Review	Classification	Date	Reference	Sequences	Attachments	Claims	KWIC	Drawi Desc
Image												

### 4. Document ID: US 6102690 A

L2: Entry 4 of 6

File: USPT

Aug 15, 2000

US-PAT-NO: 6102690

DOCUMENT-IDENTIFIER: US 6102690 A

TITLE: Recombinant organisms capable of fermenting cellobiose

DATE-ISSUED: August 15, 2000

INVENTOR-INFORMATION:

NAME CITY STATE ZIP CODE COUNTRY

Ingram; Lonnie O. Gainesville FL
Lai; Xiaokuang Gainesville FL
Moniruzzaman; Mohammed Gainesville FL
York; Sean W. Gainesville FL

US-CL-CURRENT: 431/161; 435/170, 435/189, 435/190, 435/194, 435/195, 435/252.3, 435/254.2, 435/320.1, 536/23.2

Full Title Citation Front Review Classification Date Reference Sequences Attachments KWC Draw. Desc

#### ☐ 5. Document ID: US 5916787 A

L2: Entry 5 of 6

File: USPT

Jun 29, 1999

US-PAT-NO: 5916787

DOCUMENT-IDENTIFIER: US 5916787 A

TITLE: Ethanol production in gram-positive microbes

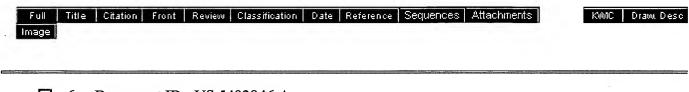
DATE-ISSUED: June 29, 1999

INVENTOR-INFORMATION:

NAME CITY STATE ZIP CODE COUNTRY

Ingram; Lonnie O'Neal Gainesville FL Barbosa-Alleyne; Maria D. F. Gainesville FL

US-CL-CURRENT: 435/161; 257/E21.317, 435/162, 435/163, 435/165, 435/252.31, 435/320.1



☐ 6. Document ID: US 5482846 A

L2: Entry 6 of 6

File: USPT

Jan 9, 1996

US-PAT-NO: 5482846

DOCUMENT-IDENTIFIER: US 5482846 A

TITLE: Ethanol production in Gram-positive microbes

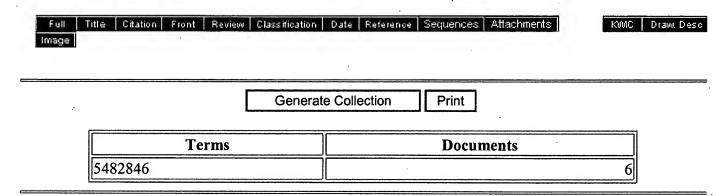
DATE-ISSUED: January 9, 1996

INVENTOR-INFORMATION:

NAME CITY STATE ZIP CODE COUNTRY

Ingram; Lonnie O'Neal Gainesville FL Barbosa-Alleyne; Maria D. F. Gainesville FL

US-CL-CURRENT: 435/161; 257/E21.317, 435/163, 435/252.31



Display Format: CIT Change Format

Previous Page Next Page

#### End of Result Set

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L2: Entry 6 of 6

File: USPT

Jan 9, 1996

US-PAT-NO: 5482846

DOCUMENT-IDENTIFIER: US 5482846 A

TITLE: Ethanol production in Gram-positive microbes

DATE-ISSUED: January 9, 1996

INVENTOR-INFORMATION:

NAME CITY STATE ZIP CODE COUNTRY

Ingram; Lonnie O'Neal Gainesville FL Barbosa-Alleyne; Maria D. F. Gainesville FL

US-CL-CURRENT: 435/161; 257/E21.317, 435/163, 435/252.31

#### CLAIMS:

#### We claim:

- 1. A Gram-positive bacterium selected from the group consisting of Bacillus subtilis and Bacillus polymyxa which has been transformed with Zymomonas mobilis genes encoding alcohol dehydrogenase and pyruvate decarboxylase, wherein said genes are expressed at sufficient levels to confer upon said Gram-positive bacterium transformant the ability to produce ethanol as a fermentation product.
- 2. A method for the production of ethanol, said method comprising:
- a) transforming a Gram-positive bacterial host selected from the group consisting of Bacillus subtilis and Bacillus polymyxa with Zymomonas mobilis genes encoding alcohol dehydrogenase and pyruvate decarboxylase, wherein said genes are expressed at sufficient levels to result in production of ethanol as a fermentation product, and
- b) growing said bacterial host so that ethanol is produced.

Generate Collection

Print

Search Results - Record(s) 11 through 20 of 37 returned.

☐ 11. Document ID: US 20020034816 A1

L5: Entry 11 of 37

File: PGPB

Mar 21, 2002

PGPUB-DOCUMENT-NUMBER: 20020034816

PGPUB-FILING-TYPE: new

DOCUMENT-IDENTIFIER: US 20020034816 A1

TITLE: Ethanol production

PUBLICATION-DATE: March 21, 2002

INVENTOR-INFORMATION:

NAME CITY STATE COUNTRY RULE-47

Green, Edward GB Surrey Javed, Muhammad Essex GB GB Baghaei-Yazdi, Namdar London

US-CL-CURRENT: <u>435/252.31</u>; <u>435/485</u>

Full	Title	Citation	Front	Review	Classification	Date	Reference	Sequences	Attachments
Image									

KWMC Drawn Desc

12. Document ID: US 20010032342 A1

L5: Entry 12 of 37

File: PGPB

Oct 18, 2001

PGPUB-DOCUMENT-NUMBER: 20010032342

PGPUB-FILING-TYPE: new

DOCUMENT-IDENTIFIER: US 20010032342 A1

TITLE: Modified ribulose 1,5-bisphosphate carboxylase/oxygenase for improvement and optimization of plant phenotypes

PUBLICATION-DATE: October 18, 2001

INVENTOR - INFORMATION:

NAME CITY COUNTRY RULE-47 STATE Stemmer, Willem P.C. Los Gatos CA US Subramanian, Venkitswaran San Diego CA US Zhu, Genhai Sunnyvale CA US Liu, Lu Redwood City CA US Selifonov, Sergey A. Los Altos CA US

US-CL-CURRENT: <u>800/298</u>; <u>435/440</u>, <u>800/278</u>

Full Title Citation Front Review Classification Date Reference Sequences Attachments
Image

KMC Draw Desc

☐ 13. Document ID: US 6541621 B1

L5: Entry 13 of 37

File: USPT

Apr 1, 2003

US-PAT-NO: 6541621

DOCUMENT-IDENTIFIER: US 6541621 B1

TITLE: Hypoxia inducible promoter

DATE-ISSUED: April 1, 2003

INVENTOR-INFORMATION:

NAME

CITY

STATE

ZIP CODE COUNTRY

Hodges; Thomas K.

West Lafayette

IN

COONIKI

Huq; Enamul

El Sobrante

CA

Hossain; Anwar

Dhaka

BD

US-CL-CURRENT: <u>536/24.1</u>; <u>435/320.1</u>

Full Title Citation Front Review Classification Date Reference Sequences Attachments

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☐ 14. Document ID: US 6485947 B1

L5: Entry 14 of 37

File: USPT

Nov 26, 2002

US-PAT-NO: 6485947

DOCUMENT-IDENTIFIER: US 6485947 B1

TITLE: Production of lactate using crabtree negative organisms in varying culture

conditions

DATE-ISSUED: November 26, 2002

INVENTOR-INFORMATION:

NAME CITY STATE ZIP CODE COUNTRY

Rajgarhia; Vineet Minnetonka MN
Hatzimanikatis; Vassily Minneapolis MN
Olson; Stacey Minneapolis MN
Carlson; Ting Dayton OH
Starr; John N. Chaska MN

Kolstad; Jeffrey J. Wayzata MN

Eyal; Aharon Jerusalem

US.-CL-CURRENT: 435/139

Full Title Citation Front Review Classification Date Reference Sequences Attachments Image

KMC Draw, Desc

IL

☐ 15. Document ID: US 6333181 B1

L5: Entry 15 of 37

File: USPT

Dec 25, 2001

US-PAT-NO: 6333181

DOCUMENT-IDENTIFIER: US 6333181 B1

TITLE: Ethanol production from lignocellulose

DATE-ISSUED: December 25, 2001

INVENTOR-INFORMATION:

NAME

CITY

STATE

ZIP CODE

COUNTRY

Ingram; Lonnie O.

Gainesville

 $\mathtt{FL}$ 

Wood; Brent E.

Gainesville

 ${ t FL}$ 

US-CL-CURRENT: 435/165; 435/163, 435/170, 435/173.2, 435/173.8, 435/277, 435/278, 435/72, 435/99

Full	Title	Citation	Front	Review	Classification	Date	Reference	Sequences	Attachments
Image									

KWMC Draws Desc

☐ 16. Document ID: US 6306639 B1

L5: Entry 16 of 37

File: USPT

Oct 23, 2001

US-PAT-NO: 6306639

DOCUMENT-IDENTIFIER: US 6306639 B1

TITLE: Genetically modified cyanobacteria for the production of ethanol, the

constructs and method thereof

DATE-ISSUED: October 23, 2001

INVENTOR-INFORMATION:

NAME

CITY

STATE

ZIP CODE

COUNTRY

Woods; Robert Paul

Markham

CA

Coleman; John Robert Deng; Ming De

Markham North York

CA

US-CL-CURRENT: 435/252.3; 536/23.1, 536/23.2, 536/24.1

Full Title Citation Front Review Classification Date Reference Sequences Attachments

KMC | Draw Desc

☐ 17. Document ID: US 6280986 B1

L5: Entry 17 of 37

File: USPT

Aug 28, 2001

US-PAT-NO: 6280986

DOCUMENT-IDENTIFIER: US 6280986 B1

TITLE: Stabilization of pet operon plasmids and <a href="ethanol production">ethanol production</a> in bacterial strains lacking lactate dehydrogenase and pyruvate formate lyase activities

DATE-ISSUED: August 28, 2001

INVENTOR - INFORMATION:

NAME CITY STATE ZIP CODE COUNTRY

Hespell; Robert B. late of Peoria ΙL Wyckoff; Herbert A. ΙL Roscoe

Dien; Bruce S. Peoria IL Bothast; Rodney J. East Peoria IL

US-CL-CURRENT: 435/161; 435/252.3, 435/252.33, 435/320.1

Front Review Classification Date Reference Sequences Attachments KWMC Draw, Desc

☐ 18. Document ID: US 6130076 A

L5: Entry 18 of 37 File: USPT Oct 10, 2000

US-PAT-NO: 6130076

DOCUMENT-IDENTIFIER: US 6130076 A

TITLE: Ethanol production using a soy hydrolysate-based medium or a yeast

autolysate-based medium

DATE-ISSUED: October 10, 2000

INVENTOR - INFORMATION:

NAME CITY STATE ZIP CODE COUNTRY

Ingram; Lonnie O. Gainesville FL

US-CL-CURRENT: 435/161

Full Title Citation Front Review Classification Date Reference Sequences Attachments KOMC Draw, Desc

□ 19. Document ID: US 6107093 A

L5: Entry 19 of 37 File: USPT Aug 22, 2000

US-PAT-NO: 6107093

DOCUMENT-IDENTIFIER: US 6107093 A

TITLE: Recombinant cells that highly express chromosomally-integrated heterologous

genes

DATE-ISSUED: August 22, 2000

INVENTOR - INFORMATION:

NAME CITY STATE ZIP CODE COUNTRY

Ingram; Lonnie O. Gainesville FLOhta; Kazuyoshi Gainesville FI. FL

Wood; Brent E. Gainesville US-CL-CURRENT: 435/440; 257/E21.317, 435/252.3, 435/252.33, 536/23.7, 536/24.1

KWMC Draw, Desc Review Classification Date Reference Image ☐ 20. Document ID: US 6102690 A L5: Entry 20 of 37 File: USPT Aug 15, 2000 US-PAT-NO: 6102690 DOCUMENT-IDENTIFIER: US 6102690 A TITLE: Recombinant organisms capable of fermenting cellobiose DATE-ISSUED: August 15, 2000 INVENTOR-INFORMATION: NAME CITY STATE ZIP CODE COUNTRY Ingram; Lonnie O. Gainesville FLGainesville Lai; Xiaokuang FLMoniruzzaman; Mohammed Gainesville FLGainesville York; Sean W. FL US-CL-CURRENT: 431/161; 435/170, 435/189, 435/190, 435/194, 435/195, 435/252.3, 435/254.2, 435/320.1, 536/23.2 Citation Front Review Classification Date Reference Sequences Attachments KWWC Drawl Desc **Generate Collection** Print **Terms Documents** 37 L4 and (gene or dna)

Display Format: - Change Format

Previous Page Next Page

**Generate Collection** 

Print

**Search Results** - Record(s) 21 through 30 of 37 returned.

☐ 21. Document ID: US 5932456 A

L5: Entry 21 of 37

File: USPT

Aug 3, 1999

US-PAT-NO: 5932456

DOCUMENT-IDENTIFIER: US 5932456 A

TITLE: Production of ethanol and other fermentation products from biomass

DATE-ISSUED: August 3, 1999

INVENTOR-INFORMATION:

NAME

CITY

STATE

ZIP CODE

COUNTRY

Van Draanen; Arlen

Bellevue

WA

Mello; Steven

Bedford

NH

US-CL-CURRENT: 435/144; 435/157, 435/160, 435/161, 435/163, 435/165, 435/171, 435/267

Full Title Citation Front Review Classification Date Reference Sequences Attachments Image

KMMC Draw, Desc

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☐ 22. Document ID: US 5916787 A

L5: Entry 22 of 37

File: USPT

Jun 29, 1999

US-PAT-NO: 5916787

DOCUMENT-IDENTIFIER: US 5916787 A

TITLE: Ethanol production in gram-positive microbes

DATE-ISSUED: June 29, 1999

INVENTOR-INFORMATION:

NAME

CITY

STATE ZIP CODE

COUNTRY

Ingram; Lonnie O'Neal

Gainesville

FL FL

Barbosa-Alleyne; Maria D. F.

Gainesville

US-CL-CURRENT: <u>435/161</u>; <u>257/E21.317</u>, <u>435/162</u>, 43<u>5/163</u>, 435/165, 435/252.31, 435/320.1

Full Title Citation Front Review Classification Date Reference Sequences Attachments
Image

KWMC Draw. Desc

☐ 23. Document ID: US 5821093 A

L5: Entry 23 of 37

File: USPT

Oct 13, 1998

US-PAT-NO: 5821093

DOCUMENT-IDENTIFIER: US 5821093 A

TITLE: Recombinant cells that highly express chromosomally-integrated heterologous

genes

DATE-ISSUED: October 13, 1998

INVENTOR-INFORMATION:

NAME CITY STATE ZIP CODE COUNTRY

Ingram; Lonnie O. Gainesville FL
Ohta; Kazuyoshi Gainesville FL
Wood; Brent E. Gainesville FL

 $\text{US-CL-CURRENT: } \underline{435/161}; \ \underline{257/E21.317}, \ \underline{435/163}, \ \underline{435/252.3}, \ \underline{435/252.33}, \ \underline{536/23.2}, \\ \underline{ 536/23.2}, \ \underline{ 536/23.2}, \ \underline{ 536/23.2}, \\ \underline{ 536/23.2}, \ \underline{ 536/23.2}, \ \underline{ 536/23.2}, \\ \underline{ 536/23.2}, \ \underline{ 536/23.2}, \ \underline{ 536/23.2}, \\ \underline{ 536/23.2}, \ \underline{ 536/23.2}, \ \underline{ 536/23.2}, \\ \underline{ 536/23.2}, \ \underline{ 536/23.2}, \ \underline{ 536/23.2}, \\ \underline{ 536/23.2}, \ \underline{ 536/23.2}, \ \underline{ 536/23.2}, \\ \underline{ 536/23.2}, \ \underline{ 536/23.2}, \ \underline{ 536/23.2}, \\ \underline{ 536/23.2}, \ \underline{ 536/23.2}, \ \underline{ 536/23.2}, \\ \underline{ 536/23.2}, \ \underline{ 536/23.2}, \ \underline{ 536/23.2}, \\ \underline{ 536/23.2}, \ \underline{ 536/23.2}, \ \underline{ 536/23.2}, \\ \underline{ 536/23.2}, \ \underline{ 536/23.2}, \ \underline{ 536/23.2}, \\ \underline{ 536/23.2}, \ \underline{ 536/23.2}, \ \underline{ 536/23.2}, \\ \underline{ 536/23.2}, \ \underline{ 536/23.2}, \ \underline{ 536/23.2}, \\ \underline{ 536/23.2}, \ \underline{ 536/23.2}, \ \underline{ 536/23.2}, \\ \underline{ 536/23.2}, \ \underline{ 536/23.$ 

<u>536/23.7</u>, <u>536/24.1</u>

Full Title Citation Front Review Classification Date Reference Sequences Attachments KMC Draw. Desc

☐ 24. Document ID: US 5677154 A

L5: Entry 24 of 37 File: USPT Oct 14, 1997

US-PAT-NO: 5677154

DOCUMENT-IDENTIFIER: US 5677154 A

\*\* See image for Certificate of Correction \*\*

TITLE: Production of ethanol from biomass

DATE-ISSUED: October 14, 1997

INVENTOR-INFORMATION:

NAME CITY STATE ZIP CODE COUNTRY

Van Draanen; Arlen Haverhill MA Mello; Steven Bedford NH

US-CL-CURRENT: 435/163; 435/161, 435/165, 435/171, 435/267

Full Title Citation Front Review Classification Date Reference Sequences Attachments KMC Draw Desc

☐ 25. Document ID: US 5607672 A

L5: Entry 25 of 37 File: USPT Mar 4, 1997

US-PAT-NO: 5607672

DOCUMENT-IDENTIFIER: US 5607672 A

TITLE: Replacement therapy for dental caries

DATE-ISSUED: March 4, 1997

INVENTOR - INFORMATION:

NAME

CITY

STATE ZIP CODE COUNTRY

Hillman; Jeffrey D.

Gainesville

FL

US-CL-CURRENT: 424/50; 424/93.44, 435/252.3

Full Title Citation Front Review Classification Date Reference Sequences Attachments Image

KMMC Draw, Desc

26. Document ID: US 5554520 A

L5: Entry 26 of 37

File: USPT

Sep 10, 1996

US-PAT-NO: 5554520

DOCUMENT-IDENTIFIER: US 5554520 A

TITLE: Ethanol production by recombinant hosts

DATE-ISSUED: September 10, 1996

INVENTOR-INFORMATION:

NAME

CITY

STATE

ZIP CODE

COUNTRY

Fowler; David E.

Gainesville

FL

Horton; Philip G.

Gainesville

FL

Ben-Bassat; Arie

Gainesville

FL

US-CL-CURRENT: 435/165; 257/E21.317, 435/162

Title Citation Front Review Classification Date Reference Seguences Attachments

KWMC - Draw, Desc

Image

☐ 27. Document ID: US 5487989 A

L5: Entry 27 of 37

File: USPT

Jan 30, 1996

US-PAT-NO: 5487989

DOCUMENT-IDENTIFIER: US 5487989 A

TITLE: Ethanol production by recombinant hosts

DATE-ISSUED: January 30, 1996

INVENTOR-INFORMATION:

NAME CITY

ZIP CODE

COUNTRY

Fowler; David E.

Gainesville

FL

STATE

Horton; Philip G. Ben-Bassat; Arie

Gainesville Gainesville FL

US-CL-CURRENT: 435/165; 257/E21.317, 435/162, 435/209, 435/252.3, 435/852

Title Citation Front Review Classification Date Reference Sequences Attachments

KMMC | Draww Desc

28. Document ID: US 5482846 A

L5: Entry 28 of 37

File: USPT

Jan 9, 1996

US-PAT-NO: 5482846

DOCUMENT-IDENTIFIER: US 5482846 A

TITLE: Ethanol production in Gram-positive microbes

DATE-ISSUED: January 9, 1996

INVENTOR-INFORMATION:

NAME CITY STATE ZIP CODE COUNTRY

Ingram; Lonnie O'Neal Gainesville FL Barbosa-Alleyne; Maria D. F. Gainesville FL

US-CL-CURRENT: 435/161; 257/E21.317, 435/163, 435/252.31

Full Title Citation Front Review Classification Date Reference Sequences Attachments

KWMC | Drawi Desc

☐ 29. Document ID: US 5424202 A

L5: Entry 29 of 37

File: USPT

Jun 13, 1995

US-PAT-NO: 5424202

DOCUMENT-IDENTIFIER: US 5424202 A

TITLE: Ethanol production by recombinant hosts

DATE-ISSUED: June 13, 1995

INVENTOR-INFORMATION:

NAME CITY STATE ZIP CODE COUNTRY

Ingram; Lonnie O. Gainesville FL
Beall; David S. Gainesville FL
Burchhardt; Gerhard F. H. Gainesville FL

Guimaraes; Walter V. Vicosa BR Ohta; Kazuyoshi Miyazaki JP

Wood; Brent E. Gainesville FL Shanmugam; Keelnatham T. Gainesville FL

US-CL-CURRENT: <u>435/161</u>; <u>257/E21.317</u>, <u>435/165</u>, <u>435/252.3</u>, <u>435/320.1</u>, <u>435/847</u>, <u>435/854</u>

| Full | Title | Citation | Front | Review | Classification | Date | Reference | Sequences | Attachments | KMC | Draw. Desc | Image |

☐ 30. Document ID: US 5270175 A

L5: Entry 30 of 37 . File: USPT Dec 14, 1993

US-PAT-NO: 5270175

DOCUMENT-IDENTIFIER: US 5270175 A

TITLE: Methods and compositions for producing metabolic products for algae

DATE-ISSUED: December 14, 1993

INVENTOR-INFORMATION:

NAME

CITY

STATE

ZIP CODE

COUNTRY

Moll; Benjamin A.

Berkeley

CA

US-CL-CURRENT: 435/41; 435/161, 435/320.1, 435/410, 435/69.1, 435/70.1, 435/946,

536/23.2

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L5: Entry 27 of 37

File: USPT

Jan 30, 1996

US-PAT-NO: 5487989

DOCUMENT-IDENTIFIER: US 5487989 A

TITLE: Ethanol production by recombinant hosts

DATE-ISSUED: January 30, 1996

INVENTOR-INFORMATION:

NAME CITY STATE ZIP CODE COUNTRY

Fowler; David E. Gainesville FL Horton; Philip G. Gainesville FL Ben-Bassat; Arie Gainesville FL

US-CL-CURRENT: 435/165; 257/E21.317, 435/162, 435/209, 435/252.3, 435/852

#### CLAIMS:

What is claimed is:

- 1. A method for producing ethanol from cellulose-containing biomass, comprising the steps of:
- A. contacting, in a first reaction vessel, said biomass with a polysaccharase such that cellulose in said biomass is broken down into simpler oligosaccharides and/or monosaccharides,

wherein said contacting is carried out at a temperature of from about 40.degree. C. to about 60.degree. C. and a pH of from about 4.5 to about 5.0;

- B. producing from said first reaction vessel a sugar solution comprising said simpler oligosaccharides and/or monosaccharides;
- C. introducing said sugar solution into a fermentor which comprises gram-negative enteric recombinant host microorganisms capable of fermenting said simpler oligosaccharides and/or monosaccharides into ethanol; and
- D. fermenting said simpler oligosaccharides and/or monosaccharides into ethanol at a temperature of from about 30.degree. C. to about 35.degree. C. and a pH of about 6.0,

wherein said microorganism is capable of fermenting monosaccharides into ethanol, and comprises a recombinant host, other than Escherichia coli, comprising a first heterologous <u>DNA</u> coding for <u>alcohol dehydrogenase and pyruvate decarboxylase</u>, wherein said heterologous <u>DNA is from Zymomonas mobilis</u> and wherein said host expresses said heterologous <u>DNA</u> at a sufficient functional level so as to facilitate the production of ethanol as the primary fermentation product by said host,

wherein said host also produces a polysaccharase, and said host further comprises a second heterologous <u>DNA</u> segment, the expression product of which is said polysaccharase.

- 2. A method according to claim 1, wherein a first stream from said fermentor is withdrawn and used to cool the sugar solution produced in Step B.
- 3. A method according to claim 2, wherein said first stream is introduced into said first vessel after cooling said sugar solution produced in Step B.
- 4. A method according to claim 1, wherein the sugar solution produced in Step B is passed through an ultrafiltration unit having an upper molecular weight cut-off ultrafiltration membrane to obtain an ultrafiltration product solution and a second solution,

said ultrafiltration product solution comprising predominantly molecules having a molecular weight below the molecular weight cut-off of said ultrafiltration membrane, said product solution comprising at least some of said oligosaccharides and/or monosaccharides obtained from Step B, and

said second solution comprising predominantly molecules having a molecular weight above the molecular weight cut-off of said ultrafiltration membrane.

- 5. A method according to claim 4, wherein said product solution predominantly comprises molecules having a molecular weight of less than about 25,000.
- 6. A method according to claim 4, wherein the ultrafiltration product solution is subjected to reverse osmosis to obtain a first stream comprising predominantly water and a second stream comprising at least some of the oligosaccharides and/or monosaccharides.
- 7. A method according to claim 6, wherein said stream comprising predominantly water is recycled to the first reaction vessel.
- 8. A method according to claim 1, wherein the sugar solution produced in Step B is subjected to reverse osmosis to obtain a first stream comprising predominantly water and a second stream comprising at least some of the oligosaccharides and/or monosaccharides.
- 9. A method according to claim 8, wherein said stream comprising predominantly water is recycled to the first reaction vessel.
- 10. A method according to claim 1, wherein the contacting in said first reaction vessel is carried out at a temperature of from about 50.degree. C. to about 60.degree. C.
- 11. A method according to claim 1, wherein the contacting in said first reaction vessel is carried out at a temperature of from about 50.degree. C. to about 55.degree. C.
- 12. A method according to claim 1, wherein enzymes which break down cellulose into simpler oligosaccharides and/or monosaccharides are added to said fermentor.
- 13. The method of claim 1, wherein said host is selected from the group consisting of Erwinia, Klebsiella and Xanthomonas.
- 14. The method of claim 13, wherein said host is selected from the group consisting of Erwinia and Klebsiella.
- 15. The method of claim 14, wherein said host is Klebsiella oxytoca M5A1(pLOI555), ATCC 68564, deposited Mar. 14, 1991.
- 16. The method of claim 1, wherein said host has been transformed with a plasmid comprising genes coding for alcohol dehydrogenase and pyruvate decarboxylase, wherein said host expresses said genes to produce alcohol dehydrogenase and pyruvate decarboxylase at a sufficient functional level to facilitate the production of ethanol as the primary fermentation product by said host.
- 17. The method of claim 16, wherein said plasmid comprises Zymomonas mobilis

genes coding for alcohol dehydrogenase and pyruvate decarboxylase.

- 18. The method of claim 16, wherein said plasmid further comprises a lac promoter which directs the expression of said genes coding for alcohol dehydrogenase and pyruvate decarboxylase.
- 19. The method of claim 16, wherein said plasmid has been designated pLOI555.
- 20. The method of claim 1, wherein said polysaccharase is a cellulolytic enzyme.
- 21. The method of claim 20, wherein said polysaccharase is selected from the group consisting of an endoglucanase, cellobiohydrolase, .beta.-glucosidase, and .beta.-glucanase.
- 22. The method of claim 21, wherein polysaccharase is an expression product of a celD gene.
- 23. The method of claim 22, wherein said celD <u>gene</u> is derived from Clostridium thermocellum.
- 24. The method of claim 21, wherein said polysaccharase is at least partially secreted by said host.
- 25. The method of claim 21, wherein said polysaccharase is accumulated in said host.
- 26. A method for producing ethanol from cellulose-containing biomass, comprising the steps of:
- A. contacting, in a first reaction vessel, said biomass with a polysaccharase such that oligosaccharide in said biomass is broken down into simpler oligosaccharides and/or monosaccharides,
- wherein said contacting is carried out at a temperature of from about 40.degree. C. to about 60.degree. C. and a pH of from about 4.5 to about 5.0;
- B. producing from said first reaction vessel a sugar solution comprising said simpler oligosaccharides and/or monosacchrides;
- C. introducing said sugar solution into a fermentor which comprises gram-negative enteric recombinant host microorganisms capable of fermenting said simpler oligosaccharides and/or monosaccharides into ethanol; and
- D. fermenting said simpler oligosaccharides and/or monosaccharides into ethanol at a temperature of from about 30.degree. C. to about 35.degree. C. and a pH of about 6.0,

wherein said microorganism is capable of fermenting both monosaccharides and oligosaccharides into ethanol, and said microorganism further comprises a recombinant host comprising first heterologous <u>DNA from Zymomonas mobilis</u> coding for <u>alcohol dehydrogenase and pyruvate decarboxylase</u>, respectively, wherein said host

- (A) further comprises <u>genes</u> coding for proteins which enable said host to transport and metabolize an oligosaccharide, and
- (B) expresses said  $\underline{\text{genes}}$  and said heterologous  $\underline{\text{DNA}}$  at a level such that ethanol is produced as the  $\underline{\text{primary}}$  fermentation product  $\underline{\text{by}}$  said host from the metabolism of said oligosaccharide, and

wherein said recombinant host also produces a polysaccharase, and said host further comprises a second heterologous  $\underline{\text{DNA}}$  segment, the expression product of which is said polysaccharase.

27. The method of claim 26, wherein said recombinant host is selected from the

group consisting of Erwinia and Klebsiella.

- 28. The method of claim 27, wherein said recombinant host is Klebsiella oxytoca M5A1(pLOI555), ATCC 68564, deposited Mar. 14, 1991.
- 29. The method of claim 26, wherein said oligosaccharide is selected from the group consisting of dimers and trimers.
- 30. The method of claim 26, wherein said polysaccharase is a cellulolytic enzyme.
- 31. The method of claim 30, wherein said polysaccharase comprises the expression product of a cellulase gene of Cellulomonas fimi, and said host secretes at least some of said polysaccharase.
- 32. The method of claim 26, wherein said polysaccharase is selected from the group consisting of an endoglucanase, cellobiohydrolase, .beta.-glucosidase, .beta.-glucanase and hemicellulase, arabinosidase.
- 33. The method of claim 32, wherein said polysaccharase is an expression product of a celD  $\underline{\text{gene}}$ .
- 34. The method of claim 33, wherein said celD  $\underline{\text{gene}}$  is derived from Clostridium thermocellum.
- 35. The method of claim 26, wherein said host further comprises an additional heterologous <u>DNA</u> segment, the expression product of which is a protein involved in the transport of mono- and/or oligosaccharides into the recombinant host.
- 36. The method of claim 26, wherein said polysaccharase is at least partially secreted by said host.
- 37. The method of claim 26, wherein said polysaccharase is accumulated in said host.
- 38. The method of claim 37, wherein said host further comprises an additional heterologous  $\underline{DNA}$  segment, the expression product of which is an additional polysaccharase that is at least partially secreted by said host.
- 39. The method of claim 38, wherein said additional polysaccharase comprises the expression product of a cellulase gene of Cellulomonas fimi.

# **WEST Search History**

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COST IN U.S. DOLLARS

SINCE FILE ENTRY

TOTAL SESSION

FULL ESTIMATED COST

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FILE 'MEDLINE' ENTERED AT 10:45:30 ON 25 JUL 2003

FILE 'CAPLUS' ENTERED AT 10:45:30 ON 25 JUL 2003

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FILE 'EMBASE' ENTERED AT 10:45:30 ON 25 JUL 2003

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FILE 'SCISEARCH' ENTERED AT 10:45:30 ON 25 JUL 2003 COPYRIGHT 2003 THOMSON ISI

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=> focus 13

PROCESSING COMPLETED FOR L3

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5 FILES SEARCHED...

29 L4 AND 1988-1995/PY

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ANSWER 1 OF 29

MEDLINE on STN

ACCESSION NUMBER:

91282482 MEDLINE

DOCUMENT NUMBER:

PubMed ID: 2059047 91282482

TITLE:

Genetic improvement of Escherichia coli for ethanol

production: chromosomal integration of

Zymomonas mobilis genes encoding

pyruvate decarboxylase and

alcohol dehydrogenase II.

AUTHOR: CORPORATE SOURCE: Ohta K; Beall D S; Mejia J P; Shanmugam K T; Ingram L O Department of Microbiology and Cell Science, University of

Florida, Gainesville 32611.

CONTRACT NUMBER:

GM37403 (NIGMS)

SOURCE:

APPLIED AND ENVIRONMENTAL MICROBIOLOGY, (1991 Apr)

57 (4) 893-900.

Journal code: 7605801. ISSN: 0099-2240.

PUB. COUNTRY:

United States

DOCUMENT TYPE:

Journal; Article; (JOURNAL ARTICLE)

LANGUAGE:

English

FILE SEGMENT:

Priority Journals

ENTRY MONTH:

199107

ENTRY DATE:

Entered STN: 19910818

Last Updated on STN: 20000303 Entered Medline: 19910729

AB Zymomonas mobilis genes for pyruvate

decarboxylase (pdc) and alcohol dehydrogenase

II (adhB) were integrated into the Escherichia coli chromosome within or near the pyruvate formate-lyase **gene** (pfl). Integration improved the stability of the Z. mobilis genes in E. coli, but further selection was required to increase expression. Spontaneous mutants were selected for resistance to high level of chloramphenicol that also expressed high levels of the Z. mobilis genes. Analogous mutants were selected for increased expression of **alcohol** 

dehydrogenase on aldehyde indicator plates. These mutants were functionally equivalent to the previous plasmid-based strains for the fermentation of xylose and glucose to ethanol. Ethanol concentrations of 54.4 and 41.6 g/liter were obtained from 10% glucose and 8% xylose, respectively. The efficiency of conversion exceeded theoretical limits (0.51 g of ethanol/g of sugar) on the basis of added sugars because of the additional production of ethanol from the catabolism of complex nutrients. Further mutations were introduced to inactivate succinate production (frd) and to block homologous recombination (recA).

L5 ANSWER 2 OF 29 CAPLUS COPYRIGHT 2003 ACS on STN

ACCESSION NUMBER:

1995:992753 CAPLUS

DOCUMENT NUMBER:

124:28129

TITLE:

Ethanol production with

recombinant Gram-positive microbes expressing

exogenous pyruvate decarboxylase and alcohol dehydrogenase genes

INVENTOR(S):

Ingram, Lonnie O'Neal; Barbosa-Alleyne, Maria de F. S.

PATENT ASSIGNEE(S):

University of Florida, USA

SOURCE:

PCT Int. Appl., 33 pp. CODEN: PIXXD2

DOCUMENT TYPE:

Patent

LANGUAGE:

English

FAMILY ACC. NUM. COUNT:

PATENT INFORMATION:

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APPLICATION NO. DATE
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                        A1 19951012
     WO 9527064
                                               WO 1995-US4012 19950330 <--
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              RU, SG, SI, SK, TJ, TT, UA, UZ, VN
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PRIORITY APPLN. INFO.:
                                             US 1988-239099 B2 19880831
                                             US 1989-352062 A2 19890515
                                             US 1990-624227 B2 19901207
                                             US 1991-670821 B2 19910318
                                             US 1992-846344 A2 19920306
                                             US 1992-946290 A2 19920917
                                             US 1993-260517 A2 19930305
                                             WO 1995-US4012
                                                               W 19950330
```

AB The subject invention concerns the transformation of Gram-pos. bacteria with heterologous genes which confer upon these microbes the ability to produce ethanol as a fermn. product. Specifically exemplified is the transformation of bacteria with genes, obtainable from Zymomonas mobilis, which encode pyruvate decarboxylase and alc. dehydrogenase. A recombinant Bacillus subtilis expressing Z. mobilis pdc and adhB genes was created.

ANSWER 3 OF 29 CAPLUS COPYRIGHT 2003 ACS on STN

1994:433306 CAPLUS ACCESSION NUMBER:

DOCUMENT NUMBER:

121:33306

TITLE:

Processes for ethanol production

INVENTOR(S):

Fowler, David E.; Horton, Philip G.; Ben-Bassat, Arie

PATENT ASSIGNEE(S):

Bioenergy International, 1.c., USA

SOURCE:

PCT Int. Appl., 172 pp.

CODEN: PIXXD2

DOCUMENT TYPE:

Patent

LANGUAGE:

English

10

FAMILY ACC. NUM. COUNT:

PATENT INFORMATION:

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                                     19940331
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                                                                           W 19930917
```

AB Novel plasmids comprising genes which code for alc. dehydrogenase and pyruvate decarboxylase are described. Also described are recombinant hosts which have been transformed with genes coding for alc. dehydrogenase and pyruvate decarboxylase. By virtue of their transformation with these genes, the recombinant hosts are capable of producing significant amts. of ethanol as a fermn. product. Also disclosed are methods for increasing the growth of recombinant hosts and methods for reducing the accumulation of undesirable metabolic products in the growth medium of these hosts. Also disclosed are recombinant hosts capable of producing significant amts. of ethanol as a fermn. product of oligosaccharides and plasmids comprising genes encoding polysaccharides, in addn. to the genes described above which code for alc. dehydrogenase and pyruvate decarboxylase. Further, methods are described for producing ethanol from oligomeric feedstock using the recombinant hosts described above. Also provided is a method for enhancing the prodn. of functional proteins in a recombinant host comprising overexpressing an adhB gene in the host. Further provided are process designs for fermenting oligosaccharide-contg. biomass to ethanol.

ANSWER 4 OF 29 CAPLUS COPYRIGHT 2003 ACS on STN

ACCESSION NUMBER:

1992:649986 CAPLUS

DOCUMENT NUMBER:

117:249986

TITLE:

**Ethanol production** by by bacteria carrying foreign genes for alcohol

dehydrogenase and pyruvate

decarboxylase

INVENTOR(S):

Ingram, Lonnie O.; Beall, David S.; Burchhardt,

Gerhard F. H.; Guimaraes, Walter V.; Ohta, Kazuyoshi; Wood, Brent E.; Shanmugam, Keelnatham T.; Fowler,

David A.; Ben-Bassat, Arie

PATENT ASSIGNEE(S):

University of Florida, USA; Bioenergy International,

L.C.

PCT Int. Appl., 153 pp. SOURCE:

CODEN: PIXXD2

DOCUMENT TYPE:

Patent English

LANGUAGE:

FAMILY ACC. NUM. COUNT:

PATENT INFORMATION:

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                      KIND DATE
                                              APPLICATION NO. DATE
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RW: AT, BE, BF, BJ, CF, CG, CH, CI, CM, DE, DK, ES, FR, GA, GB, GN, GR, IT, LU, MC, ML, MR, NL, SE, SN, TD, TG
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PRIORITY APPLN. INFO.:
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                                          US 1988-239099 B2 19880831
                                          US 1989-352062 A2 19890515
                                          US 1990-624277
                                                           B2 19901207
                                          WO 1992-US1807
                                                            A 19920318
```

AB Bacterial hosts, excluding Escherichia coli, expressing heterologous genes for alc. dehydrogenase (I) and pyruvate decarboxylase (II) are used for manuf. of EtOH. II is used to prevent accumulation of acid metabolites. Plasmids, e.g. pLOI555 carrying genes for I and II of Zymomonas mobilis driven by the lac promoter, are provided for prepn. of the host. The method is further improved by transforming the host with genes for proteins that facilitate transport and metab. of oligosaccharides, e.g., of C5-6 sugars, which host is, preferably, also expressing a heterologous gene for a polysaccharase such as a cellulolytic enzyme, a xylanolytic enzyme, or a starch-degrading enzyme. These hosts also preferably express heterologous genes for polysaccharide- degrading enzymes (e.g. those degrading cellulose, xylans, or starch). A cost-effective fermn. process for manufg. EtOH from oligosaccharide feedstocks using a single, genetically engineered microorganism is also disclosed. An ethanologenic strain Klebsiella oxytoca M5A1(pLOI555) was prepd. and was further transformed with plasmid pLOI2003 encoding xylanase (gene xynZ) and xylosidase (gene xylB) of Clostridium thermocellum to obtain a transformant capable of converting xylan to EtOH.

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ANSWER 5 OF 29 CAPLUS COPYRIGHT 2003 ACS on STN
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ACCESSION NUMBER:

1992:19704 CAPLUS

DOCUMENT NUMBER:

116:19704

TITLE:

Metabolic engineering of Klebsiella oxytoca M5A1 for

ethanol production from xylose and

qlucose

AUTHOR(S):

Ohta, Kazuyoshi; Beall, D. S.; Mejia, J. P.;

Shanmugam, K. T.; Ingram, L. O.

CORPORATE SOURCE:

Dep. Microbiol. Cell Sci., Univ. Florida, Gainesville,

FL, 32611-0100, USA

SOURCE:

Applied and Environmental Microbiology (1991

), 57(10), 2810-15

CODEN: AEMIDF; ISSN: 0099-2240

DOCUMENT TYPE: Journal LANGUAGE: English

The efficient diversion of pyruvate from normal fermentative pathways to ethanol prodn. in K. oxytoca M5A1 requires the expression of

Zymomonas mobilis genes encoding both pyruvate

decarboxylase and alc. dehydrogenase. Final ethanol concns.

obtained with the best recombinant, strain M5A1 (pLOI555), were in excess of 40 g/L with an efficiency of 0.48 g of ethanol (xylose) and 0.50 g of ethanol (glucose) per g of sugar, as compared with a theor. max. of 0.51 q ethanol/q sugar. The maximal volumetric productivity per h for both sugars was 2.0 g/L. This volumetric productivity with xylose is almost twice that previously obtained with ethanologenic Escherichia coli. Succinate was also produced as a minor product during fermn.

ANSWER 6 OF 29 CAPLUS COPYRIGHT 2003 ACS on STN

ACCESSION NUMBER:

1991:534238 CAPLUS

DOCUMENT NUMBER:

115:134238

TITLE:

Ethanol production using

recombinant mutant Escherichia coli Ingram, Lonnie O.; Clark, David P.

INVENTOR(S): PATENT ASSIGNEE(S):

University of Florida, USA

SOURCE:

U.S., 5 pp. Cont.-in-part of U.S. Ser. No. 239,099,

abandoned. CODEN: USXXAM

DOCUMENT TYPE: LANGUAGE:

Patent English

10

FAMILY ACC. NUM. COUNT:

PATENT INFORMATION:

APPLICATION NO. DATE PATENT NO. KIND DATE ----------US 5028539 A 19910702 US 1988-2/40, \_\_\_\_ US 1988-239099 B2 19880831 US 1988-274075 19881121 <--PRIORITY APPLN. INFO.:

Escherichia coli with mutations which result in alc. dehydrogenase

hyperprodn. are transformed with the Zymomonas mobilis

pyruvate decarboxylase gene. These
microorganisms can be used to manuf. EtOH. The recombinant mutants grew to higher cell d. than did the parent strain. The increase in d. and the extent to which EtOH accumulated in the medium correlated with the level of expression of the decarboxylase.

ANSWER 7 OF 29 CAPLUS COPYRIGHT 2003 ACS on STN

ACCESSION NUMBER:

1990:422259 CAPLUS

DOCUMENT NUMBER:

113:22259

TITLE:

Ethanol production by genetically engineered Escherichia coli strains

INVENTOR(S):

Ingram, Lonnie O.; Conway, Tyrrell; Alterthum, Flavio

PATENT ASSIGNEE(S): University of Florida, USA

SOURCE:

PCT Int. Appl., 37 pp.

CODEN: PIXXD2

DOCUMENT TYPE:

Patent

LANGUAGE:

English

FAMILY ACC. NUM. COUNT: 10

PATENT INFORMATION:

PAT	TENT NO.		KIND DATE		APPLICATION NO.	DATE
		<b>-</b>				
WO	9002193		A1 19900308		WO 1989-US3753	19890829 <
	W: BG,	BR,	HU, JP, KP, KR,	RO,	SU	
	RW: AT,	BE,	CH, DE, FR, GB,	IT,	LU, NL, SE	
US	5000000		A 19910319		US 1989-352062	19890515 <
EΡ	431047		A1 19910612		EP 1989-909966	19890829 <
	R: AT,	BE,	CH, DE, FR, GB,	IT,	LI, LU, NL, SE	
HU	60328		A2 19920828		HU 1989-5771	19890829 <

JP 05502366 T2 19930428 JP 1989-509287 19890829 <--CA 1989-609829 19890830 <--CA 1335430 A1 19950502 PRIORITY APPLN. INFO.: US 1988-239099 A 19880831 US 1989-352062 A 19890515 WO 1989-US3753

AB DNA fragments comprising alc. dehydrogenase and pyruvate decarboxylase (enzymes of the ethanologenic pathway) genes were isolated from Zymomonas mobilis and cloned in E. coli cells using different plasmids as expression vehicles. Both enzymes were expressed at high level and this resulted in elevated EtOH productivity. Using 12% glucose, 12% lactose, and 8% xylose as substrates, the EtOH yields in E. coli were 58, 52, and 42 g/L, resp., or 95, 80, and 102%, resp., of theor.

L5 ANSWER 8 OF 29 BIOSIS COPYRIGHT 2003 BIOLOGICAL ABSTRACTS INC. on STN

ACCESSION NUMBER: 1993:73335 BIOSIS DOCUMENT NUMBER: PREV199395037835

TITLE: Use of the tac promoter and lacI-q for the controlled

expression of Zymomonas mobilis

fermentative genes in Escherichia coli and

Zymomonas mobilis.

AUTHOR(S): Arfman, N.; Worrell, V.; Ingram, L. O. (1)

CORPORATE SOURCE: (1) Dep. Microbiol. Cell Sci., University Florida,

Gainesville, Fla. 32611

SOURCE: Journal of Bacteriology, (1992) Vol. 174, No. 22, pp.

7370-7378.

ISSN: 0021-9193.

DOCUMENT TYPE: Article LANGUAGE: English

AB The Zymomonas mobilis genes encoding alcohol dehydrogenase I (adhA), alcohol dehydrogenase

II (adhB), and pyruvate decarboxylase (pdc) were

overexpressed in Escherichia coli and Z. mobilis by using a broad-host-range vector containing the tac promoter and the lacI-q

repressor gene. Maximal IPTG (isopropyl-beta-D-

thiogalactopyranoside) induction of these plasmid-borne genes in Z.

mobilis resulted in a 35-fold increase in alcohol

dehydrogenase I activity, a 16.7-fold increase in alcohol dehydrogenase II activity, and a 6.3-fold increase in

dehydrogenase II activity, and a 6.3-fold increase in pyruvate decarboxylase activity. Small changes in the

activities of these enzymes did not affect glycolytic flux in cells which are at maximal metabolic activity, indicating that flux under these conditions is controlled at some other point in metabolism. Expression of adhA, adhB, or pdc at high specific activities (above 8 IU/mg of cell protein) resulted in a decrease in glycolytic flux (negative flux control coefficients), which was most pronounced for pyruvate decarboxylate. Growth rate and flux are imperfectly coupled in this organism. Neither a twofold increase in flux nor a 50% decline from maximal flux caused any immediate change in growth rate. Thus, the rates of biosynthesis and growth in this organism are not limited by energy generation in rich medium.

L5 ANSWER 9 OF 29 BIOSIS COPYRIGHT 2003 BIOLOGICAL ABSTRACTS INC. on STN

ACCESSION NUMBER: 1988:481687 BIOSIS

DOCUMENT NUMBER: BA86:112997

TITLE: ALCOHOL PRODUCTION FROM GLUCOSE AND XYLOSE USING

ESCHERICHIA-COLI CONTAINING ZYMOMONAS-

MOBILIS GENES.

AUTHOR(S): NEALE A D; SCOPES R K; KELLY J M

CORPORATE SOURCE: DEP. BIOCHEMISTRY, LA TROBE UNIV., BUNDOORA, VICTORIA 3083,

AUSTRALIA.

SOURCE: APPL MICROBIOL BIOTECHNOL, (1988) 29 (2-3), 162-167.

CODEN: AMBIDG. ISSN: 0175-7598.

FILE SEGMENT: BA; OLD LANGUAGE: English

AB An Escherichia coli strain containing a recombinant plasmid encoding the pyruvate decarboxylase and alcohol

dehydrogenase genes from Zymomonas mobilis

metabolized glucose and xylose to near theoretical yields of ethanol. Enzyme activity measurements indicate high expression levels of both plasmid-encoded Zymomonas proteins in the recombinant E. coli. The expression in E. coli is under the control of a promoter in the Zymomonas sequence upstream of the pyruvate decarboxylase

gene. The maximum ethanol level, using 4% glucose as substrate, was 1.8% (w/v) in anaerobic conditions. In aerobic conditions the natural repression of E. coli alcohol dehyrogenase results in less ethanol production from clones expressing only Zymomas pyruvate

decarboxylase.

L5 ANSWER 10 OF 29 BIOSIS COPYRIGHT 2003 BIOLOGICAL ABSTRACTS INC. on STN

ACCESSION NUMBER: 1988:154499 BIOSIS

DOCUMENT NUMBER: BA85:78152

TITLE: EXPRESSION OF DIFFERENT LEVELS OF ETHANOLOGENIC ENZYMES

FROM ZYMOMONAS-MOBILIS IN RECOMBINANT

STRAINS OF ESCHERICHIA-COLI.

AUTHOR(S): INGRAM L O; CONWAY T

CORPORATE SOURCE: DEP. MICROBIOLOGY AND CELL SCIENCE, UNIVERSITY FLORIDA,

GAINESVILLE, FLA. 32611.

SOURCE: APPL ENVIRON MICROBIOL, (1988) 54 (2), 397-404.

CODEN: AEMIDF. ISSN: 0099-2240.

FILE SEGMENT: BA; OLD LANGUAGE: English

AB The expression of Zymomonas mobilis genes encoding

pyruvate decarboxylase and alcohol

dehydrogenase II in Escherichia coli converted this organism from the production of organic acids to the production of ethanol. Ethanol was produced during both anaerobic and aerobic growth. The extent to which these ethanologenic enzymes were expressed correlated with the extent of ethanol production. The replacement of organic acids with ethanol as a metabolic product during aerobic and anaerobic growth resulted in dramatic increases in final cell density, indicating that these acids (and the associated decline in pH) are more damaging than the

production of ethanol. Of the plasmids examined, the best plasmid for

growth and ethanol production expressed

pyruvate decarboxylase and alcohol
dehydrogenase II at levels of 6.5 and 2.5 IU/mg of total cell
protein, respectively.

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L6 1 L4 AND 1984-1987/PY

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L6 ANSWER 1 OF 1 MEDLINE on STN ACCESSION NUMBER: 88105387 MEDLINE

DOCUMENT NUMBER: 88105387 PubMed ID: 3322191

TITLE: Genetic engineering of ethanol production

in Escherichia coli.

AUTHOR: Ingram L O; Conway T; Clark D P; Sewell G W; Preston J F CORPORATE SOURCE: Department of Microbiology and Cell Science, University of

Florida, Gainesville 32611.

SOURCE: APPLIED AND ENVIRONMENTAL MICROBIOLOGY, (1987 Oct)

53 (10) 2420-5.

Journal code: 7605801. ISSN: 0099-2240.

PUB. COUNTRY: United States

DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)

LANGUAGE: English

FILE SEGMENT: Priority Journals

ENTRY MONTH: 198802

ENTRY DATE:

Entered STN: 19900305

Last Updated on STN: 19980206 Entered Medline: 19880202

AB The genes encoding essential enzymes of the fermentative pathway for ethanol production in Zymomonas

mobilis, an obligately ethanologenic bacterium, were inserted into Escherichia coli under the control of a common promoter. Alcohol dehydrogenase II and pyruvate decarboxylase

from Z. mobilis were expressed at high levels in E. coli, resulting in increased cell growth and the production of ethanol as the principal fermentation product from glucose. These results demonstrate that it is possible to change the fermentation products of an organism, such as E. coli, by the addition of genes encoding appropriate enzymes which form an alternative system for the regeneration of NAD+.

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(FILE 'HOME' ENTERED AT 10:44:44 ON 25 JUL 2003)

FILE 'MEDLINE, CAPLUS, BIOSIS, EMBASE, BIOTECHDS, SCISEARCH' ENTERED AT 10:45:30 ON 25 JUL 2003

L1 104 S ALCOHOL DEHYDROGENASE AND PYRUVATE DECARBOXYLASE AND (DNA OR L2 72 DUP REM L1 (32 DUPLICATES REMOVED)

L2 72 DUP REM L1 (32 DUPLICATES REMOVED)
L3 55 S L2 AND ZYMOMONAS MOBILIS

L4 55 FOCUS L3 1-

L5 . 29 S L4 AND 1988-1995/PY L6 1 S L4 AND 1984-1987/PY

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COST IN U.S. DOLLARS	SINCE FILE	TOTAL
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FULL ESTIMATED COST	59.36	59.57
DISCOUNT AMOUNTS (FOR QUALIFYING ACCOUNTS)	SINCE FILE	TOTAL
	· ENTRY	SESSION
CA SUBSCRIBER PRICE	-3.91	-3.91

STN INTERNATIONAL LOGOFF AT 10:53:51 ON 25 JUL 2003